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Molecular cloning of cDNA for human prostatic acid phosphatase.

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A human liver cDNA library in lambda gt11 was screened with polyclonal antiserum to human acid phosphatase isoenzyme 2a/4. About eleven positive clones have been obtained. Two clones, lambda Hap21 and lambda Hap22 were further characterized: clone lambda Hap21 contained a 0.8-kb cDNA insert and clone lambda Hap22 a 1.8-2.0-kb insert. XbaI digestion of lambda Hap22 generated two fragments of 1.0 and 0.9 kb. BglII digestion resulted in a 1.2-kb fragment and several smaller fragments of undetermined size. Clone lambda Hap22 contained all the genes carried by lambda gt11 (lac5cI857nin5Sam100) and the 2-kb insert. An Escherichia coli(lambda Hap22) lysogen was generated, and its acid phosphatase activity was approximately ten-fold higher than that in the control nonlysogenic lysate. Western-blot analysis of total proteins present in this E. coli(lambda Hap22) lysate revealed that the non-induced lambda Hap22 prophage directed the synthesis of an approx. 175-kDa protein. This protein was recognized by antibody to the human acid phosphatase isoenzyme 2a/4 and anti-beta-galactosidase and was produced only upon induction with IPTG. These results indicated that lambda Hap22 carried a major portion of the gene coding for the human acid phosphatase isoenzyme 2a and/or 4 and this protein fragment of acid phosphatase was sufficient to manifest enzymatic activity.

PMID: 2965059 [PubMed - indexed for MEDLINE]

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